

ラット脳シナプトソームにおけるキョートルフィン (Tyrosine—Arginine) 合成酵素

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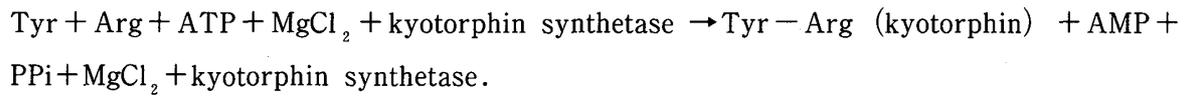
Kyotorphin (Tyrosine—Arginine) Synthetase in Rat Brain Synaptosomes

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ABSTRACT: Kyotorphin (Tyr-Arg) is a unique neuropeptide which produces analgesia by releasing Met-enkephalin from slices of the brain and spinal cord. Recent studies revealed that kyotorphin possesses the properties of neurotransmitter/neuroregulator. In the present study, we identified a kyotorphin synthetase in the soluble fraction of rat brain synaptosomes (synaptosol) and characterized it. The enzyme partially purified with Sephacryl S-300 showed an absolute requirement for ATP, MgCl₂, tyrosine, and arginine. The optimal pH was 7.5–9.0 and the pI was determined to be 6.1–6.2 by isoelectric focusing. The K_m was 25.6 μM for tyrosine, 926 μM for arginine, 294 μM for ATP, and 442 μM for MgCl₂. The V_{max} was 34.0 pmol/mg of protein/h. The apparent molecular size of this “Kyotorphin synthetase” further purified by the DE52 column was 240,000–245,000 daltons, estimated using TSKgel G4000SW column chromatography. The enzyme reaction is represented by the following equation: Tyr + Arg + ATP + MgCl₂ + kyotorphin synthetase → Tyr-Arg (kyotorphin) + AMP + P_i + MgCl₂ + Kyotorphin synthetase. The regional distribution and subcellular localization of the synthetase showed a close correlation to that of kyotorphin levels in the rat brain. The amounts of kyotorphin formed from amino acids by the synthetase in the dialyzed synaptosol was 3.0–4.0 times higher than that from precursor proteins by processing enzymes within the 30 min incubation.

抄録 本研究において我々は、ラット脳シナプトソームの可溶性画分（シナプトソール）に於てキョートルフィン合成酵素を同定し、その特性を検討した。キョートルフィン合成酵素の分子サイズは、240,000-245,000ダルトンであった。酵素反応は次式の通りである

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この合成酵素のラット脳内分布及び細胞下局在は、キョートルフィン含量のそれらと密接な相関性を示した。30分以内のインキュベーションでは、シナプトソールにおけるアミノ酸からの合成酵素によるキョートルフィンの生成量は、プロセシング酵素による前駆体蛋白からの生成量よりも3-4倍高かった。

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